lines 5-12. Support for the deletion of element (a) can be found at least in claim 1 as filed. Support for "individual microspheres" having a triphasic release profile is inherent in the description of the triphasic release profile of the microspheres demonstrated throughout the specification. Support for individual microspheres having a triphasic release profile can also be found at least at page 45, lines 43-47, where it is stated that "the microspheres are preferably designed to produce an in vitro second burst at the same time." Support for the term "in vitro" can similarly be found at least at page 45, lines 43-47 as well as from page 29, line 27 to page 30, line 2. Support for the terms "first antigen burst phase," "second slow release phase," and "third antigen burst phase" can be found at least at page 23, lines 20-22. Further support for the amendments is set forth below. Claim 28 is added; support can be found in claim 1 as filed. No new matter is added.

Drawings

The Examiner's maintenance of the objections to the drawings is acknowledged. A submission of Formal Drawings accompanies this Preliminary Amendment.

Informalities

The Examiner objected to claim 1, lines 7 and 10, for the repetition of "about" and to an improper recitation of claim 1 in the last amendment. Applicants have deleted the repetition of "about" from line 10 and amended line 7 above. As amended above, claim 1 recites that "the microspheres have a median diameter of about 20 to 100 μm".

Rejections Under 35 U.S.C. § 112, First Paragraph

Claims 1, 4-9, and 23-27 were rejected under 35 U.S.C. § 112, first paragraph, for failure to satisfy the description requirement. Specifically, the Examiner objected to the terms: "1 milliliter of aqueous antigen per 3 grams of polymer or less;" "about 1 to 2 days" with reference to the length of the initial burst of antigen release; and "beginning at the completion of." These rejections are moot insofar as these phrases have been eliminated by amendment above.

Although Applicants believe that the specification provides ample support for the deleted language, Claim 1 has been amended in an effort to expedite prosecution. The antigen release phases are now defined as a first antigen burst phase, a second slow release phase, and a third antigen burst phase. The phases are further defined by length in days, by when the phase occurs in relation to suspension of the microspheres in a release medium, and by the amount of antigen released. Assays for determining *in vitro* microsphere release are taught in the specification. Furthermore, daily microsphere release is exemplified in Figure 8 of the application (see the histograph along the baseline which uses the scale on the right of the figure). Three phases of release can be discerned in the histograph.

In the first antigen burst phase, antigen is released from the microspheres "over a period of about three days after suspension of the microspheres in a release medium." This is consistent with the release histograph shown in Figure 8, where it can be seen that antigen is released during the first three days after suspension, after which time the antigen release drops to a basal level. Suitable in vitro release media for such assays are described at page 30, lines 1-2, and at page 45, lines 45-47.

The claims also now recite that the second slow release phase occurs after the first antigen burst phase, and that the third antigen burst phase occurs after the second slow release phase. Occurrence of the second slow release phase after the first antigen burst phase, and of the third antigen burst phase after the second slow release phase is supported throughout the specification. For example, the specification states at page 21, lines 28-31: "Typically, an antigen of interest will be formulated in PLGA microspheres to provide a desired period of time *between* the first and second bursts of antigen and to provide a desired amount of antigen in each burst." Page 5, lines 30-32, of the specification states: "The microspheres of the instant invention release the antigen and/or adjuvant in three phases: an initial burst, a slow release, and a second burst."

The second slow release phase extends "from about the fourth to at least about the thirtieth day after suspension wherein the daily release of antigen from the microspheres is less than in the first antigen burst phase or a third antigen burst phase." This timing is supported by the histograph in Figure 8, where the daily release of antigen from the suspended microspheres

drops to a basal level on the fourth day after suspension and remains at a low basal level until at least day 30.

In the third antigen burst phase, "antigen is released from the microspheres at a rate of greater than 10 percent per week during a period of from about seven to about 30 days starting from about 30 to about 180 days after suspension." Support for release of greater than 10 percent of the antigen per week for at least one week can be found in footnote "b" to Table 6 of the application (page 48), and second burst times of up to 30 days are shown in the Table. Support for the third antigen burst phase starting from about 30 to about 180 days after suspension can be found in Table 6, Figure 8, and at page 46, line 16, through page 47, line 14.

Because the specification fully supports the language of the pending claims, the claims are asserted to satisfy section 112, first paragraph.

Rejections Under 35 U.S.C. § 103

Claims 1, 4-9 and 23-27 stand rejected under 35 U.S.C. § 103 as detailed below. These rejections are traversed. The individual rejections under § 103 are each addressed in turn.

Requirements for establishing a *prima facie* case of obviousness. "To establish a prima facie case of obviousness, three basic criteria must be met. First, there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references when combined) must teach or suggest all the claim limitations. The teaching or suggestion to make the claimed combination and the reasonable expectation of success must both be found in the prior art, and not based on applicant's disclosure. *In re Vaeck*, 947 F.2d 488, 20 USPQ2d 1438 (Fed. Cir. 1991)." MPEP 2142.

The rejection over Sanders et al., Eldridge et al., and Jeffery et al. The Examiner rejected claims 1, 4-9, and 23-27 under § 103 over Sanders et al., J. Pharmaceutical Sciences 73:1294-97 (1984) ("Sanders"), in view of Eldridge et al., Mol. Immunology 28:287-94 (1991)

("Eldridge"), and further in view of Jeffery et al., Pharmaceutical Research 10:362-68 (1993) ("Jeffery"). The rejection is respectfully traversed.

Sanders was cited for teaching microspheres having an alleged triphasic release profile (Office Action 6/23/98, pages 10-11). Eldridge was cited as teaching "the use of PLGA as a safe vaccine delivery system," and Jeffery was cited as disclosing encapsulation methods using a variety of aqueous antigen volumes. *Id.* at 12.

Sanders teaches the use of PLGA microspheres to provide the controlled release of nafarelin acetate to suppress estrus in rats and rhesus monkeys. Nafarelin is a small-molecule therapeutic agent that acts as an analog of luteinizing hormone-releasing hormone. It has been used to inhibit ovulation in humans and to treat endometriosis. It is not desirable to receive a subtherapeutic dose of agent in either the inhibition of ovulation or in treating endometriosis. Nor is it desirable for the agent to be an antigen, as the immune response of the patient to the agent would suppress its therapeutic effect.

Sanders does not teach or suggest that triphasic release was desirable, nor does Sanders teach or suggest the incorporation of an antigen. Sanders was not even convinced that triphasic release was demonstrated, stating that the compound release profile shown in Fig. 2 "exhibited complex kinetic behavior, *possibly* reflecting a triphasic release mechanism." Page 1296, second column, emphasis added. Figure 2 itself shows that Sanders' "triphasic kinetics" profile is conveniently drawn *directly* through their data points when a zero-order kinetics line could have been drawn within their standard deviations. Indeed, the *in vivo* tests of those same spheres showed what was clearly a monophasic release of compound in the Rhesus monkey in Figure 3(b).

The Examiner stated that Sanders discloses a 69:31 coplymer that gives a triphasic compound release over 90 days (Office Action 3/17/99, page 4-5). Sanders does not teach the molecular weight of their polymers, which were prepared in-house (p. 1294) and are thus not available to the public. Applicants have shown that the source of PLGA can dramatically affect the properties of microspheres made from PLGA (see Examples 1-2C at pages 25-49, particularly Tables 2, 3, 5 and 6, comparing the properties of microspheres made from PLGA from

Boehringer Ingelheim and Medisorb Technologies International L.P.). Sanders does not teach which component is present at 69 parts, and which at 31. Sanders does not teach how their microspheres were dried, which Applicants have shown dramatically affects the release profile of microspheres. Furthermore, there is no reasonable likelihood that a larger antigen would be released from Sanders' microspheres in the same manner as Sanders' small-molecule therapeutic agent. Sanders is therefore not enabling.

Furthermore, Sanders does not teach or suggest a triphasic release profile having a first antigen burst phase where "about 0.5 to 30 percent of the antigen is released from the microspheres over a period of about three days after suspension of the microspheres in a release medium" or a second slow release phase "extending from about the fourth to at least about the thirtieth day after suspension wherein the daily release of antigen is less than in the first antigen burst phase or a third antigen burst phase." Assuming, *arguendo*, that Sanders does show a triphasic release profile, the first release phase of Sanders' small molecule apparently continues at least until day 10 in Figure 4. Similarly, assuming Sanders demonstrates a second slow release phase, Sanders' second slow release phase does not start at about day 4 in Figure 4, nor does it extend to at least about the thirtieth day in Figure 2. Sanders therefore does not teach or suggest microspheres meeting the claimed first antigen burst phase or second slow release phase. Nor does Sanders enable any way of achieving individual microspheres having those release characteristics.

Eldridge teaches the injection of 1-10 μm PLGA microspheres containing approximately 1% by weight of a formalinized toxoid vaccine of staphylococcal enterotoxin B. Eldridge teaches that these can be "blended" with similar 20-50 μm (variously described as 20-125 μm) microspheres and injected to induce both a primary and secondary immune response. Microspheres of <10 μm were reported to be rapidly phagocytized after injection, presumably providing an initial antigen burst. The 20-50 μm microspheres were presumably included to give a later antigen burst. The timing of this burst can be inferred from Eldridge as follows. Antibodies against the antigen encapsulated in 20-50 μm microspheres were not detected until 30 days after injection of the microspheres, and antibody titer peaked at a low level 50-60 days post-injection (Eldridge, page 290). The antibody titer in a primary immune response generally rises

about 10-14 days after exposure to antigen. Therefore Eldridge's 20-50 μm microspheres released their antigen at about 16-20 days post-injection. Whereas Claim 1 recites a "second slow release phase . . . extending . . . to at least the thirtieth day after suspension," followed by an antigen burst, the corresponding burst in Eldridge's blended preparation occurred earlier than day 30 after injection. Thus, even with a blended population of microspheres, Eldridge did not enable a triphasic release profile within the scope of Applicants' claims, as neither the second slow release phase nor the third antigen burst phase occurred with the claimed timing. Nor does Eldridge provide motivation for any other timing.

Moreover, the triphasic release profile of Claim 1 can be achieved without using blended populations of microspheres such as those described by Eldridge. In response to this distinction, the Examiner pointed out that the claims do not recite that the microspheres are "non-blended" (Office Action, 3/17/99, page 5, end of first paragraph). Claim 1 has been amended to reflect that "individual microspheres have an *in vitro* antigen release profile characterized by three phases." The claim thus emphasizes that it is Applicants' individual microspheres that have a triphasic release profile, but does not exclude the possibility that such microspheres could be blended with other microspheres having different release profiles, as taught in the specification.

Jeffery teaches only the use of PLGA polymers of 1-2 μm in diameter and was cited by the Examiner for the "incorporation of antigens in poly(lactide-co-glycolide) systems," with the Examiner conceding that Jeffery does not teach triphasic release. Office Action 3/17/99, page 5. Jeffery additionally used microspheres of a small size outside the scope of Applicants' claims. Applicants have demonstrated that alteration of production parameters of PLGA microspheres leads to dramatically different release characteristics, and Eldridge and Jeffery both showed that particles as small as those used by Jeffery are rapidly phagocytized and degraded so that triphasic release would not be possible from such microspheres.

As none of the cited references teach or suggest individual microspheres comprising an antigen and having a triphasic release profile, *prima facie* obviousness of the claimed invention has not been established.

Furthermore, none of the references teach or suggest microspheres having a first antigen burst phase wherein about 0.5 to 30 percent of the antigen is released from the microspheres during the first three days. Nor do any of the references teach or suggest microspheres having a second slow release phase extending from about the fourth to at least about the thirtieth day after suspension wherein the daily release of antigen is less than that of the first or third antigen burst phases. As none of the references teach, suggest or enable microspheres with these characteristics, *prima facie* obviousness cannot be established.

Moreover, the record is devoid of any specific motivation to make the claimed combination of references. The Court of Appeals for the Federal Circuit has forcefully stated that an Examiner must provide a specific motivation in the art for combining elements from cited art in order to establish obviousness of the combination.

> "[C]ase law makes clear that the best defense against the subtle but powerful attraction of a hindsight-based obviousness analysis is rigorous application of the requirement for a showing of the teaching or motivation to combine prior art references. ... Combining prior art references without evidence of such a suggestion, teaching, or motivation simply takes the inventor's disclosure as a blueprint for piecing together the prior art to defeat patentability--the essence of hindsight. ... [Evidence of a suggestion, teaching, or motivation to combine] must be clear and particular. ... Broad conclusory statements regarding the teaching of multiple references, standing alone, are not 'evidence.' ... [If] a reference-by-reference, limitation-by-limitation analysis fails to demonstrate how the [cited] references teach or suggest their combination ... to yield the claimed invention, . . . a conclusion of obviousness based on such an analysis as a matter of law, cannot stand." *In re Dembiczak*, 175 F.3d 994, 999, 1000, 50 USPQ2d 1614, 1617, 1618 (Fed. Cir. 1999), emphasis added.

No clear, particular suggestion or motivation in the prior art to make the specific combination of an antigen with individual microspheres having the specific claimed triphasic release profile has been provided by the Examiner. Only Eldridge suggests the desirability of polyphasic release of an antigen, but Eldridge teaches that this is accomplished by mixing microspheres having different release profiles outside the scope of Applicants' claims. Eldridge's suggestion of a different way to accomplish polyphasic release does not provide motivation for making Applicants' individual microspheres exhibiting a specific triphasic release profile. In the absence of such specific suggestions in the prior art, a rejection identifying

individual elements of the claimed combination is based merely on hindsight in light of Applicants' disclosure.

Accordingly, withdrawal of the rejection is respectfully requested.

The rejection over Sanders et al., Eldridge et al., Jeffery et al. and Wang et al.

Claims 5-7 were rejected under § 103 over the combination of Sanders, Eldridge, and Jeffery, in view of Wang et al., J. Controlled Release 17:23-32 (1991) ("Wang"). The rejection is respectfully traversed.

Claims 5-7 depend, directly or indirectly, from Claim 1 and recite a composition "further comprising an adjuvant." Wang was cited by the Examiner for the proposition that it was known in the art of vaccination to combine adjuvants with antigens for release from microspheres. Office Action 3/17/99, page 6, first paragraph. The microspheres of Claim 1 have been distinguished above. Wang does not provide a teaching or suggestion of a triphasic release profile meeting Applicants' claim limitations that would remedy that deficiency in the primary references. The Examiner noted as much in the Office Action, stating that "Wang is not directed to triphasic release pattern" (page 6, first paragraph).

Wang used Carbopol 951 in an attempt to enhance protein loading because Carbopol can act as a gelling agent similar to the gelatin used by Ogawa et al. (p. 29, first column, stating: "This study employed a variation on [Ogawa's] approach using Carbopol 951 at elevated pH *to gel the inner phase*" [emphasis added]). Wang's incorporation of Carbopol produced microspheres that "showed a high burst effect of 40-50%" of the incorporated protein and gave high continuous release such that 65-75% of the protein was released within the first 25 days (p. 28, Figure 4 and second column, second paragraph). Wang noted that Carbopol may be acting "as a wetting agent or may promote the osmotic uptake of water into the microsphere's pore structure resulting in a large burst effect" (page 29, second column).

Wang's formulation thus produced microspheres having continuous rather than triphasic release of protein. An attempt to use a (water-in-oil)-in-water emulsion with Carbopol produced microspheres with an even greater initial burst (60%; see Figure 5 and p. 29, second column). Not only did Carbopol make the initial burst larger than recited in Applicants' claims, it caused

the continued release of BSA from the microspheres so that no second slow release phase wherein the daily release of antigen is lower than in the first or third antigen burst phases occurred. In fact, there was no third antigen burst phase at all.

Thus, the cited references do not teach or suggest all the claim elements. Further, as there was no specific motivation in the art to modify or combine the references to produce a composition comprising an adjuvant and microspheres having the claimed release profile and no reasonable expectation of success in doing so based on the teachings of Wang, *prima facie* obviousness has not been established. The Examiner is therefore respectfully requested to withdraw this rejection.

The rejection over Sanders et al., Eldridge et al., Jeffery et al., Wang et al. and Newman et al. Claim 8 was rejected under § 103 over the combination of Sanders, Eldridge, Jeffery, and Wang in view of Newman et al., AIDS Research and Human Retroviruses 8:1413-18 (1992) ("Newman"). The rejection is respectfully traversed.

Claim 8 depends from Claim 5, and ultimately therefore from Claim 1. Claim 8 recites a composition "wherein the adjuvant is QS21." Newman is cited simply for its teaching that QS21 "is non-toxic and augments both antibody responses and cell-mediated immunity." Office Action 6/23/98, page 15.

The rejection is traversed on two grounds. First, microspheres comprising an antigen and having the claimed triphasic release profile are not obvious, as set forth above, and therefore a composition comprising such microspheres and QS-21 cannot be obvious. Second, none of the references suggest the *specific* combination of QS-21 with such microspheres, as is required by Federal Circuit precedent. Newman does not mention microspheres; none of the other references mentions QS-21.

Thus, the cited references do not teach or suggest all the claim elements, there was no motivation in the art to modify or combine the references to produce a composition comprising QS-21 and microspheres having the claimed release profile, and there was no reasonable expectation of success in doing so. As *prima facie* obviousness has not been established, the Examiner is therefore respectfully requested to withdraw this rejection.

The rejection over Floy et al. The Examiner rejected Claims 1, 4, 9, and 23-27 under 35 U.S.C. 103 over Floy et al. ("Design of Biodegradable Polymer Systems for Controlled Release of Bioactive Agents," Polymeric Delivery Systems: Properties and Applications, American Chemical Society, Washington, D.C., 1993) ("Floy"). The rejection is respectfully traversed.

Floy is a review article discussing the release of therapeutic agents, not antigens, from polymeric delivery systems. Floy present no new data; in fact, Floy cites Sanders regarding triphasic release. Floy neither teaches nor suggests the specific elements of the triphasic release profile of Applicants' claimed composition. It is a fundamental requirement of *prima facie* obviousness that all the claim elements be taught or suggested in the cited art. As Floy does not suggest use of an antigen or the specific elements of the triphasic release profile of Applicants' composition, not all the claim elements are provided by Floy. No *prima facie* case of obviousness has therefore been established.

The Examiner is respectfully requested to withdraw the rejection.

The rejection over Floy et al. and Immunization Practices Advisory Committee Recommendations. The Examiner rejected Claims 5-7 under 35 U.S.C. 103 as unpatentable over Floy in view of Immunization Advisory Committee, Clin. Pharm. 8:839-851 (1989) ("the IAC recommendations"). The rejection is respectfully traversed.

Floy does not teach or suggest any antigen, or combining an antigen with microspheres, much less microspheres having the claimed release profile. Floy does not teach or suggest adding an adjuvant to microspheres. The IAC recommendations do not teach or suggest the use of microspheres with adjuvants or antigens, much less microspheres having the claimed release profile. There is no specific motivation in the art to combine these references, and the references do not teach or suggest the elements of the triphasic release profile of Applicants' claimed microspheres. And there was no reasonable expectation that individual microspheres could release antigen with the triphasic release profile of the claimed composition. Therefore *prima facie* obviousness has not been established. Withdrawal of the rejection is respectfully requested.

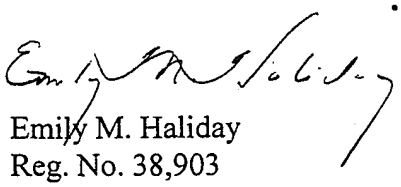
The rejection over Floy et al., Immunization Practices Advisory Committee Recommendations and Newman et al. The Examiner rejected Claim 8 under 35 U.S.C. § 103 over Floy in view of the IAC recommendations, and further in view of Newman. The rejection is respectfully traversed. As discussed above, Newman is cited solely for its disclosure of the QS21 adjuvant. Neither Floy nor the IAC recommendations nor Newman suggest combining microspheres with antigens, nor do the references suggest microspheres having the claimed release profiles. As there is no motivation in the art to combine the references and no reasonable expectation of success in doing so, *prima facie* obviousness has not been established. Withdrawal of the rejection is respectfully requested.

Conclusion

Applicants respectfully submit that the application is now in condition for allowance. Accordingly, a notice of allowance is requested. If the Examiner has any questions regarding this submission, the Examiner is requested to contact the undersigned at (640) 849-4908.

Date: August 7, 2000

Respectfully submitted,



Emily M. Haliday
Reg. No. 38,903

Skjerven, Morrill, MacPherson, LLP
25 Metro Drive, Suite 700
San Jose, CA 95110
Telephone: (408) 487-1296
Facsimile: (408) 453-7979

648408 v1